THE ANATOMY OF THE DIASPININE SCALE INSECT EPIDIASPIS PIRICOLA (DEL GUER.).

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The anatomy of several species of scale insects (Coccidæ) has been studied in the entomological laboratories of Stanford University, these species representing several different genera, such as Physokermes, Ceroputo and Icerya. All these, however, are of more or less generalized type and show but little marked divergence from a common form. Comparatively little work has been done in this laboratory or elsewhere on the anatomy of the more specialized Coccidæ, the Diaspinæ.

It is the purpose of this paper to describe the more important anatomical characteristics that are representative of the sub-family Diaspinæ as a whole. A knowledge of the facts of the make-up and functions of the parts should add to the interest of any study of the sub-family, whether the student have the viewpoint of an economic or systematic entomologist. In taking a particular member of the sub-family for this study, I have chosen the species familiarly known as the Italian pear scale *Epidiaspis piricola* (Del Guer). It is one commonly found about the University and in the whole Santa Clara Valley.

In studying the anatomy of such a small and well chitinized insect a number of difficulties of technic present themselves. The first thing that must be considered is the method of killing. Embedding and cutting were also features that demanded considerable experimentation before desirable results were obtained.

Three killing fluids were experimented with in particular; Towers' formula No. 2, Gilson's, and hot water. The latter proved to be the most useful. A fourth, Carnoy's mixture of six parts of absolute alcohol, three parts of chloroform and one part of glacial acetic acid, was also used somewhat for killing. It brought out the nervous system admirably, but the other tissues were badly distorted or destroyed by the action of this powerful agent.

The orienting of the material in the paraffine, ready for sectioning, was a little problem in itself, on account of the minuteness of the insect. The best results were obtained by the following means: A small paper receptacle was half filled with melted 55% paraffine and allowed to cool considerably. The insect was then taken up in melted paraffine in a warm pipette, and dropped into the receptacle, which was then filled. The specimen was then moved into desired position with a warmed needle. After following this method of embedding, little trouble was experienced in trimming up the block, and the cutting could be done with accuracy in the plane wished.

Necessary care must, of course, be taken in seeing that all material is thoroughly embedded or the sections are very liable to tear when the knife comes in contact with the tough, chitinous covering of the insect. This chitin was found to be extremely impenetrable, and all attempts at staining *in toto* proved futile, though specimens were allowed to stay in the staining fluid for days.

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NERVOUS SYSTEM.

The nervous system (Plate XII, Fig. 4) of the Coccidæ seems to vary little in structural characteristics in the many widely differing groups of the family. The central system consists of a fused, bi-lobed cephalic ganglion forming the brain, and a prominent compressed thoracic ganglion from which four pairs of lateral nerves are given off. The posterior pair curve out towards the lateral margins of the insect and then curve back again apparently fusing or at least giving off a great number of smaller nerves which form a delicate fan-shaped nerve center in the pygidium. The circum-oesophageal connectives, (Plate XII, Fig. 4-a) are exceptionally long in Epidiaspis and, together with this lesser nerve center just mentioned, represent the chief differences of the Diaspinæ from the other forms studied. This pushing forward of the brain and the lengthening of the oesophageal commissures is undoubtedly a result of specialization arising from the development of the mouth-parts. The extremely well developed muscles that govern the sucking apparatus are found just below the brain, and it is undoubtedly their growth that has pushed forward the more or less functional brain. However, a nerve can be seen arising from either side of the lobes reaching out towards the antennal rudiments.

Experiments were undertaken to ascertain the insect's sensitiveness to touch, which show that there is a decided lack of visible response to any sort of stimulus. The only movement that could be noted was that of the drawing in, or telescoping, of the posterior region when touched with a needle. This shortening takes place through the contraction of the segments. No other movement of the body was observed in response to other stimulants such as light, heat and water.

DIGESTIVE SYSTEM.

The Diaspinæ present an extraordinary arrangement of the digestive system, diverging in this respect considerably from the other sub-families. Dr. A. Berlese, the well known Italian biologist, seems to be about the only man who has done any considerable amount of work on this group, and he reports a very novel condition of the system. He describes elaborately in his studies, the arrangement of the organs of digestion and assimilation, and finds that the stomach is entirely disconnected from the intestine and the rectum. This condition seems to be almost unbelievable. It is a condition met with usually only in certain animal forms where there occurs a regurgitation of undigestible foods. Such an action is highly improbable among the Diaspinæ. Certainly no one has ever observed this phenomenon among them and the removal of wastes can probably be explained in another way.

The digestive epithelium of the stomach of Epidiaspis is made up of very large cells (Plate XIII, Fig. 9-a) with correspondingly large nuclei. The action of the digestive secretions on the ingested plant juices is such that it reduces them to a condition where they can be taken up by the blood-plasma and used for food, reaching this medium by osmosis through the walls of the blind sac or stomach. With the food also passes that which is of no use to the insect and which is taken care of by the exceedingly well developed Malpighian tubules, of which there is a single very large pair, (Plate XIII, Fig. 9-b). These excretory organs are fused, and at the very point of fusion a short duct leads into the rectum to which the

Malpighian tubes are attached by a filament (Plate XIII, Fig. 9-e) a short distance above the anal aperture. The proportionately large size of these tubules indicates that their function is not of ordinary or small proportions. They are made up of exceedingly large granular cells, with distinct nuclei, surrounding a thread-like lumen leading forward to the fusion of the two tubules, from which there is a connection into the tube leading into the rectum. These organs must be considered primarily as organs of excretion and capable of taking from the body cavity not only that material taken in with the food, but removing from the system the waste products of metabolism.

Dr. Berlese declares that there is absolutely no continuous connection from mouth to intestine. However, the writer finds some sections in his series that show what can hardly be denied to be direct connections, (Plate XIII, Fig. 9-c). Berlese's theory of digestion and assimilation is quite plausible and there is a good argument for its probability, for this connection, at best, is very small. The greater number of the sections that have been made—and I have sectioned several score of specimens—show the condition as Dr. Berlese describes. But it seemed possible that the result might be due to an imperfect technic, so that to find a united alimentary canal was the cause for cutting so much material. The results seem to point to this accomplishment. I think that it is in the killing that the trouble lies. We have to do with an insect with a rigid, chitinous exterior with the anal aperture and esophagus attached to chitin. The intermediate system possesses two large bodies, the stomach and the Malpighian tubules, joined by a very delicate intestine, which, unable to withstand the sudden shock of certain killing fluids, is ruptured, with the natural result that many, if not most, of the insects, show a disconnected digestive system.

This condition might possibly vary in the different Diaspinine genera, yet this is not probable. More work is still to be done on the group as a whole, and should this finding be true it will be a point of morphological importance at least, though perhaps not altering the present accepted theory in regard to the manner in which this system carries on its digestive and secretive functions.

REPRODUCTIVE SYSTEM.

The female reproductive system of Epidiaspis (Plate XIV, Figs. 14 and 15), is found to be characteristic of the usual insect type, consisting of a pair of ovaries joined to the vagina to form a figure much the shape of a capital Y. The vagina is a rather long, thick duct, lined with prominent gland cells with prominent nuclei, which undoubtedly secrete a tough, shell-like material during the passing of the eggs. The vagina opens on the ventral surface opposite the anal aperture. At the junction of the two branches of the vagina is found the minute opening of the seminal receptacle (Plate XIV, Fig. 14-b). This sperm sac is a long blind tube (Plate XIV, Fig. 16), and at the time observations were made was filled with an exceedingly large quantity of sperm cells, which could often be seen in the semi-cleared specimens under the microscope.

From the branches of the ovaries, masses of ovariole buds are given off, varying in size from a mere evagination of the egg tube to that in which the eggs are well developed (Plate XIV, Fig. 14-a). Each of these ovarioles is capable of producing a single egg, which, upon reaching maturity, passes down the slender connective into the vagina (Plate XIV, Fig. 15-c), and thence to the exterior. It is quite evident that all of these buds do not develop, and I have noted that the female apparently stops feeding to any great extent after egg laying begins. Consequently after a certain number of eggs have been deposited, she probably does not possess the vitality to bring all the others to maturity.

CIRCUMGENITAL GLANDS, OR SPINNERETS.

The study of the grouped glands or spinnerets (Plate XIV, Fig. 17 to 21), and their histology and function, offered an especial opportunity for some needed observations, and proved to be very interesting. The histology and actual function of these spinnerets have been subjects of much conjecture, although little real work seems to have been done on them. The reason for this is undoubtedly the fact that the glands (Plate XIV, Fig. 17-a) are functional for a very short period of time, and unless sections are made at this particular time, no glands can be found in connection with the grouped orifices. Sections made before the egg-laying period begins, cutting squarely through these grouped spinnerets, often show a heavily nucleated invagination of the hypoderm, but aside from this no inkling as to the function can be ascertained. Just before the insect commences egg-laying, however, a white powdery substance can be found issuing from the openings, and sections made from the material killed at this time brought out the gland cells in their minutest detail. A slender duct (Plate XIV, Fig. 17-c) is found to connect each of the circular openings with the wax secreting glands. These units vary in number of cells from one or two to six or seven, each division possessing a prominent nucleus and uniting with the main duct by a slender, thread-like lumen. Between these ducts are numerous elongate supporting cells (Plate XIV, Fig. 17-b) which have no relation with the functions of secretion, except that they may aid in keeping the passageway open.

The spinnerets are circular, rather lens-shaped, and made of chitin, through which a minute pore is found connecting the cell and duct to the exterior. This opening seems always to be uniform in its makeup—a rosette with five small parts.

Prof. E. E. Green is the author of a general rule which can be applied to Coccids possessing these glands, that they are for the most part ovo-viviparous, or egg-laying, while those that do not possess these spinnerets are viviparous. I have made observations on a number of the local species and find the rule to hold true. I have also noted that the embryo reaches a greater state of development before the egg is deposited in the cases of those species in which there are very few spinnerets, than in those that possess a large number of grouped glands. In *Aspidiotus uvae* the circumgenital glands are found in very small numbers, two or three in a group, and the species is reported to be viviparous. The opportunity of studying a large series of these insects might reveal some very interesting facts. One can easily make a mistake in examining material for this phenomenon by using adult females which have died containing well developed eggs. The eggs are not destroyed by the death of the mother, but from them hatch young, around which is the shriveled skin of the mother, and through which there is no means of escape for the newly hatched insects. Such material when mounted impresses one as being of a viviparous form. Circumstances of this nature may be the explanation of some of the seeming exceptions to the rule.

Upon the arrival of the egg-depositing season the female assumes a different posture than is its earlier position, shortening itself to a considerable extent and appearing much more rounded (Plate XIV, Fig. 21). On the insect's taking this shape the vulva opens directly back instead of on the ventral aspect. With this change comes also a shifting of the normal position of the spinnerets (Plate XIV, Fig. 19) to the position shown in Fig. 20. In Epidiaspis the spinnerets consist of five groups, three anterior and two posterior, all about equidistant from the genital aperture. This shifting from the normal, together with an enlargement and infolding of the vulva, draws the glands to a position immediately surrounding and lining the opening. The eggs passing through this opening must necessarily pass over the glands, and in this passing the moist egg picks up a quantity of the powdery secretion that has exuded, more or less covering the newly deposited eggs (Plate XIII, Fig. 21). This powdery substance not only acts as a protection to the eggs, but also aids in keeping them from drying and sticking together and thus blocking up the limited space in which the insect has to store her ova.

THE MOUTH PARTS.

The mouthparts of the Diaspinæ are, as with the other subfamilies of the Coccidæ, hard to homologize with those of other insects. They are too minute to dissect and are always flattened out of shape when mounted, thus making a detailed description of the size and shape of the various parts a difficult thing to do with accuracy. For the most part this box-like framework of the Diaspinine mouth structure can be homologized with that for other Coccidæ, as described by Putnam, Mark, Moulton and others.

This framework is a very important structure in that it serves as a means of attachment for the powerful muscles that govern the sucking and swallowing apparatus. In the main, there are two arches (Plate XIII, Fig. 11) arcus superior (*a*), and arcus inferior, (*b*), which fuse to make a very rigid structure. Just below this box-like structure is a rostrum or mentum, conical in shape and attached at the base. This conical structure is covered with a chitinous layer and inside of this are a few short muscles, which are undoubtedly used in the manipulation of the long buccal setæ, the four apparently modified mandibles and maxillæ, which are, in the case of this group of insects, extended into a long piercing beak. These four chitinous rods are arranged so that the plant juices pass up through the tube formed by their union. Attached behind the rostrum and lying free in the body cavity, is the setal pouch, extending well back towards the posterior end of the thoracic ganglion, into which the setæ are pulled when they are not in use.

At the base of the arcus inferior (Plate XIII, Fig. 11-b) is found a very interesting apparatus made particularly striking by its close resemblance to a piston valve, with all of its attachments, chamber, rod and head (Plate XIII, Fig. 12). Figure 13 shows a diagramatic cross section of this valve, showing inlet, 13-i, from salivary glands (Plate XIII, Fig. 11-d) and outlet into oesophagus 13-o.

Here again, in the impossibility of actual observation of the functioning, the interpretation of the actual function of the organ has to be based upon its structural make-up and the work that it apparently has to perform. Dr. Berlese is of the opinion that this pump is used for drawing the saliva from the large paired glands (Plate XIII, Fig. 11-e), and the arrangement of the ducts and the openings into the cylinder would seem to indicate this. Yet the rule for most insects with comparable organs is that these glands, as a result of an internal pressure caused by a continual secretion of the cells from within, or from muscular action, force the juices out. In the case of this insect no muscles are to be found that could perform this function, and, from the make-up of the glands themselves, they seem to the writer to be admirably adapted to operate through a pressure formed from within. Again, from the make-up of the long, slender, four-pieced proboscis, it would seem to be impossible to pump saliva into the plant tissues, for pressure from the inside would disrupt the tube. Necessarily, therefore, if there is a passage of fluid down this setal arrangement it would have to be done with little or no pressure. The presence of stained plant tissue at the point of puncture possibly indicates that some fluid does pass, as exemplified by the familiar reddish staining occasioned by the presence of the San Jose scale (*Aspidiotus perniciosus* Comst.)

on various fruit trees. This possibly might be explained as being the result of a mechanical stimulus and the disrupting of the cellular make-up of the plant, for the long chitinous rods that compose the mouthparts pierce and destroy a great many cells. This theory seems to be at fault however, in that all scale insects do not cause this phenomenon; for example, on the apple *A. perniciosus* causes a very distinct reddening while *E. piricola* does not.

The real function of the saliva is undoubtedly not to aid in the taking up of the plant juices, but to act upon these food properties after they have entered the insect's body. The relationship of the opening of the salivary glands to the oesophagus would seem to point to this. The food is poured into a common chamber at the base of the proboscis (Plate XIII, Fig. 12-c) passing on through the slender oesophagus (12-e) into the stomach.

The posterior part of the pharynx is a decidedly chitinized structure, apparently valvular, to which a number of powerful muscles (Fig. 12-f) are attached, and whose function it is to help force the food forward, through expansion and contraction of the walls of the oesophagus. These muscles are attached to the ventral wall of the insect, and undoubtedly act in conjunction with the large retractor muscles found directly above the pump-like cylinder of the mouthparts (Plate XIII, Fig. 12-b).

In the main the long, slender oesophagus resembles a capital letter U running forward from the chitinized pharynx parallel to the ventral wall (Plate XIII, Fig. 12-e) of the insect to about that point corresponding to the cephalic region of the chitinous box-like framework of the mouth-parts. At this point it turns toward the dorsal surface, passing between the two circum-oesophageal commissures close to the base of the brain. Here the tube turns parallel to the center, emptying into the stomach at a point just posterior to a vertical line that would run through the mentum (Plate XIII, Fig. 8). This tube is cylindrical in outline and it may be distinguished in sections from the surrounding tissue by the minute, circular cells, of which it is composed.

RESPIRATORY SYSTEM.

The Coccids possess a respiratory system that seems to be nearly uniform through the entire group, and that found in this species varies little from that of the larger representatives of the family. It consists, in Epidiaspis, of two pairs of stigmatic openings or spiracles, well guarded by hairs and spinneret-like glands which excrete a powdery wax over the exterior opening. These apertures (Plate XII, Fig. 6) possess rosettes (6-b) that make them indistinguishable from those that surround the vulva. A short tube leads from the stigma, opening into a chamber from which numerous tracheoles radiate, always, however, in a definite way so that all the individual insects of that species show the same characteristics and design (Plate VII, Fig. 2). For the most part these tubes are confined to the ventral surface, but can often be traced into the body cavity, surrounding the different groups of organs. The degree of diffusion of this system and its limit of ramification could not be determined, as only those tubes that possess a chitinization can be positively identified in the sections. In the larger, less specialized forms of Coccids, taenidial rings, the familiar characteristic of tracheal tissue are found, but in this species no such rings were noted, though a careful search for them was made. The characteristic trunk connectives joining the spiracles are very small and as a rule are unbranched. In some Coccidæ these have been found to be wanting, undoubtedly the result of degeneration.

THE CIRCULATORY SYSTEM.

The scale insects as a whole seem to be lacking in anything that may be called a definite circulatory system. No trace of a dorsal vessel can be found and no movement or pulsation of the body was noted that would indicate the presence of any such system.

EXPLANATION OF FIGURES.

PLATE XII.

Fig. 1. Pygidium of adult female, dorsal surface with circumgenital orifice and glands showing through.
Fig. 2. Respiratory system of female insect, showing two pairs of spiracles.
Fig. 3. Glandular spine, used in spinning the shell. (cross section shown in Fig. 18-e).
Fig. 4. Nervous system. a. Circumoesophageal commissures; b. brain; c. thoracic ganglion.
Fig. 5. Cross section of segments. a. chitin; b. muscle; c. muscle attachment.
Fig. 6. Spiracle. a. opening; b. wax producing spinnerets; c. tracheal tubes.

PLATE XIII.

Fig. 8. Sagittal-longitudinal section of adult female. a. stomach; b. malpighian tubules; c. rectum; d. vagina; e. ovarioles; f. thoracic ganglion; g. cephalic ganglion; h. salivary glands; i. setæ (rostral); j. setal pouch.
Fig. 9. Digestive tract. a. stomach; b. malpighian tubules; c. intestine; d. rectum; e. attachment filaments.
Fig. 10. Characteristic muscle structure found under chitinous covering.
Fig. 11. Mouth-parts and accessories. a. arcus superior; b. arcus inferior; d. valvular pump; e. salivary gland; g. costa inferior; h. costa superior; i. oesophagus (i. oesophagus drawn to one side); j. salivary ducts; k. setæ; m. muscle.
Fig. 12. Longitudinal cross section of mouth-parts. a. setæ; b. muscle; c. common chamber at base of setæ; the union point of oesophagus and duct leading from pump; d. salivary duct; e. oesophagus; f. muscles attached to base of oesophagus and to the body wall.
Fig. 13. Diagrammatic cross section of pump showing inlet and outlet. i. inlet; o. outlet.

PLATE XIV.

Fig. 14. Ovaries, dorsal view. a. ovarioles; b. seminal receptacle; c. vagina; d. vulva.
Fig. 15. Ovaries, lateral view. (lettering the same as Fig. 14).
Fig. 16. Seminal receptacle. a. sperm cells.
Fig. 17. Sagittal section of gravid female cutting through caudo and cephalolateral circumgenital glands. a. wax glands with neuclei; b. supporting cells with neuclei; c. gland ducts; d. grouped gland orifices.
Fig. 18. Cross section of spinning glands that form the shell. d. dorsal spinneret with characteristic silk gland; e. spine-like marginal spinneret; f. silk gland; g. gland found in connection with the silk gland and supposed to secrete a cement-like fluid.
Fig. 19. Normal position of the vulva and circumgenital glands of adult female. a. vulva; b. anus.
Fig. 20. Shift in position of the circumgenital glands of the adult female during egg laying.
Fig. 21. Position of adult female during egg laying. a. normal position indicated by dotted line; b. position during the depositing of the eggs.